THE JOURNAL OF INVESTIGATIVE DERMATOLOGY
Vol. 53, No. 2

Printed in U.S.A.

EPINEPHRINE INDUCTION OF WHITE HAIR IN ACI RATS*

WALTER B. SHELLEY, M.D.

AND

SVEN ÖHMAN, M.D.

ABSTRACT

A single injection of epinephrine into an area of skin of ACI rats from which pigmented hairs have been plucked leads regularly to a regrowth of completely non-pigmented hairs. The injection is effective when given any time during the 10-day anagen cycle after plucking. The longer the interval between plucking and the injection, the greater the delay in new white hair growth. The conclusion is drawn that the new white hairs reflect selective damage or destruction of the hair bulb melanocytes following the intense vasoconstriction induced by epinephrine.

Although neuro-humoral factors have been considered in the pathogenesis of graying of hair and clinicians have recorded hair turning white from fright, little experimental data appears in the literature (1, 2). We were accordingly intrigued to come upon a rather hidden observation of Findlay and Jenkinson that the single local injection of epinephrine into the skin of Ayrshire calves turned their normally brown hair white in the exact area of vasoconstriction (3). The change in color was dramatic in that the transition from brown to white in the hair shaft was sharply defined. Significantly the depigmentation was limited to the hair; the brown epidermis remained fully pigmented. The loss of hair pigment was complete and persisted for months. It occurred only after injection of epinephrine in a concentration of 1/1000; more dilute solutions were without effect. The loss of hair color was interpreted by Findlay and Jenkinson as attributable to selective destruction of the melanocyte by the intense vasoconstriction. Concomitant degenerative changes were noted in the sweat gland epithelium.

A careful search of the literature revealed only one other incidental report of the effect of epinephrine on hair pigmentation. In this study Mohn recorded that 37 black rats injected daily with epinephrine for more than 4 weeks showed a regrowth of white hair locally in plucked areas of the leg where the repeated injections were made (4). The epinephrine was administered both subcutaneously in aqueous solution and intramuscularly with an oily suspension in each instance in a dose of 0.1 mg twice daily. At times small necrotic areas of skin were noted at the site of repeated injection. White hairs appeared in the hind legs of the rats after 56 injections of epinephrine. Whitening of hairs was never noted except over the injection sites and Mohn ventured no view as to its nature, except to point out that the effect was not due to mechanical injury since the animals receiving control vehicle injections rarely grew white hairs.

No clear role for epinephrine in melanin synthesis has been discerned. Yet, Stubblefield, Escue and Utt (5) found that epinephrine did aggregate preformed melanin granules in tissue culture melanocytes derived from both mouse skin and melanomas. Such aggregation did not occur in surviving dermal melanocytes from mice (6) and the epidermal melanocytes are refractory to hormones which regularly evoke lightening of amphibian skin (7).

In the present study we have attempted to find a model for further study of the local development of achromotrichia in areas injected with epinephrine.

MATERIALS AND METHODS

Studies were done on thirty-two C57 BL/6 black mice, thirteen Brown-Norway rats, and fifty-seven ACI rats (Microbiological Associates, Inc., Bethesda, Maryland). The ACI strain was represented by inbred females, 100–150 grams in size. It has an agouti brown-black pigmented hair population on the back, whereas the ventral hair is

Supported by a John A. Hartford Foundation, Inc. grant.

Received February 24, 1969; accepted for publication March 11, 1969.

* From the Department of Dermatology, University of Pennsylvania School of Medicine, Philadelphia, Pennsylvania 19104.

white. Pigmented hairs were manually plucked from a circular area on the back. One-tenth milliliter of epinephrine solution (Parke-Davis) in 1/1000 strength (or serial dilutions thereof in sterile saline) was injected intradermally in the center of the epilated skin site. Control injections consisted of 0.9% sodium chloride, 0.1% sodium bisulfite in distilled water. A single injection generally was given each animal, but litter mates were injected on sequential days to study time relationships of the plucking, injection and regrowth. Readings of the skin color, skin appearance, hair regrowth pattern, color and density were made three times a week for six months. Representative biopsies were also taken for histologic examination.

Additional studies were made following the injection of mecholyl and nor-epinephrine, as well as local freezing.

RESULTS

None of the Brown-Norway rats nor the C57 BL/6 black mice showed any white hair formation following plucking and a single local intradermal injection of epinephrine 10^{-3} (0.1 ml for rats, 0.03 ml for mice). The injections were given to various animals on every day, 1 through 20, following plucking. In some C57 BL/6 animals two sequential injections were made, i.e., days 0 and 6, 1 and 7, 2 and 8, 3 and 9, 4 and 10, 5 and 11, 6 and 12, 7 and 13, 8 and 14, 9 and 15, 10 and 16, 15 and 21, 20 and 26. All regrew pigmented hairs.

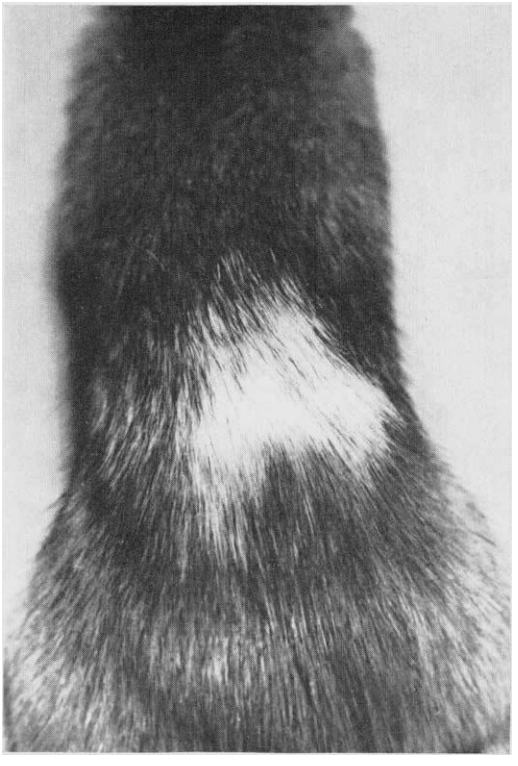

FIG. 1. *White hair growth* four weeks following injection of epinephrine (1 ml — 10^{-4}) into area 4 days after plucking an oval area of brown hairs on the back of an ACI rat.

By contrast, all of the ACI rats regrew white hair in plucked areas in which epinephrine was injected intradermally (Fig. 1). In a typical experiment, the hairs were plucked, and 0.1 ml of 10^{-3} epinephrine was injected on day 4. This was followed days later by crust formation at the injection site. The crust remained for over two weeks, to be followed by a luxuriant growth of new white hairs by the third to fourth week. Injecting the epinephrine on the day after plucking shortened the interval to regrowth of white hairs, whereas the interval could be lengthened by waiting until 10 days after plucking before injecting the epinephrine. Two injections in the same site a week apart also delayed regrowth but the same white hair appeared.

Reducing the concentration to 1/2000 made no change, but at 1/5000 the crusting was not as thick and some of the new hairs were pigmented. At a level of 1/10,000 the injection of 0.1 ml was without effect, but the whitening could be achieved by simply injecting a larger quantity, 0.25, 0.50 or 1.00 ml. At a concentration of 1/100,000 a single injection of epinephrine was followed by the regrowth of brown pigmented hairs, despite the injection of volumes up to 1.0 ml. Injection of epinephrine into unplucked skin had no effect on the color of the hair in situ, but in some instances the subsequent hair population was depigmented. Finally plucking alone was not followed by regrowth of depigmented hairs.

Norepinephrine (0.1 ml, 1/1000, 1/5000, 1/10,000) had no consistent effect on the repigmentation cycle in eight rats. It did produce crusting, but it was significantly less than that following epinephrine, and only an occasional animal showed depigmentation of hairs. Mecholyl (0.5 ml, 1/1000) was likewise without effect. Similarly the bisulfite vehicle control injections (up to 1 ml) were followed by regrowth of brown hair. Freezing the skin with dry ice for 20 and 40 seconds did induce crusting of the skin, but this was followed by regrowth of fully pigmented hairs.

Histologically the white zones showed a preponderance of melanin free hairs.

DISCUSSION

Is the public right in believing fright turns hair white? It is this question that makes the study of the effect of any stress hormone on hair pigmentation intriguing. We found that a single intradermal injection of epinephrine in a concentration as low as 1/10,000 induced the growth of nonpigmented hairs in brown ACI rats. This is in agreement with the earlier finding of Findlay and Jenkinson that epinephrine induced white hair growth in Ayrshire calves and the observation of Mohn that multiple injections of epinephrine in the leg of black rats over a period of months was followed by local depigmentation of new hair. Although there were no clues as to the possible mechanism of this striking effect from a study of the modes of action of other hormones on hair (8, 9), one might surmise that the epinephrine was acting in a chemically specific fashion, or in a nonspecific damaging way as suspected by Findlay and Jenkinson (3). Interestingly, norepinephrine did not induce the same effect as epinephrine.

To ignore basic genetic and aging determinants, hair depigmentation from the chemical standpoint may reflect either copper or vitamin deficiencies, or poisoning with phenyl thiourea, molybdenum, or chloroquin (10). The present story as we have seen it tends to be different since epinephrine fails to act in very low concentrations or at a distance, produces enough tissue necrosis to induce crust formation, and induces an irreversible effect in that subsequent hairs coming from the same follicle continue to be white. The crusting and irreversibility support the view that epinephrine may be selectively destroying or damaging the follicular melanocytes. Indeed this is compatible with the fact that damage to the skin by freezing, wounding, grafting, x-ray, irradiated RNA or non-specific means, leads to regrowth of depigmented hair in animals and man (11–18). In the case of freezing and in surgical excision followed by re-epithelialization of a wound which is kept from contracting, apparently there is actual hair neoplasia, but without reconstitution of the hair bulb melanocyte population.

The present ACI rat model provides a simple rapid method of consistently producing a population of white hairs in a pigmented laboratory animal. It should be of value for the investigation of follicular melanocyte repopulation. Further studies of the effect of repeated injections of low concentrations of epinephrine, its analogues, synergists and antagonists, should be done. In this regard it is significant that Munan (19) found that subcutaneous injections of "partially oxidized" adrenalin induced local alopecia in black rats, followed by local regrowth of gray hair which remained permanently depigmented. His report was preliminary, and he did not give details of dosage nor did he study the effect of adrenalin itself.

REFERENCES

1. Lerner, A. B.: Vitiligo. J. Invest. Derm., *32:* 285, 1959.
2. Ephraim, A. J.: On sudden or rapid whitening of the hair. Arch. Derm., *79:* 228, 1959.
3. Findlay, J. D. and Jenkinson, D. M.: Sweat gland function in the Ayrshire calf. Research in Vet. Science, *5:* 109, 1964.
4. Mohn, M. P.: The effects of different hormonal states on the growth of hair in rats, *The Biology of Hair Growth.* Eds., Montagna, W. and Ellis, R. A., Academic Press, New York, 1958.
5. Stubblefield, E., Escue, R. E. and Utt, J.: Melanin mobilization in pigment cells in the mouse. Science, *126:* 28, 1957.
6. Reams, W. M., Shervette, R. E. and Dorman, W. H.: Refractoriness of mouse dermal melanocytes to hormones. J. Invest. Derm., *50:* 338, 1968.
7. Klaus, S. N. and Snell, R. S.: The responses of mammalian melanocytes in culture to hormones. J. Invest. Derm., *48:* 352, 1967.
8. Downes, A. M. and Wallace, A. L. C.: Local effects on wool growth of intradermal injections of hormones, *Biology of the Skin and Hair Growth.* Eds., Lyne, A. G. and Short, B. F., Elsevier Publishing Co., New York, 1965.
9. Johnson, E.: Growth and replacement of hair, *Rodents in Comparative Physiology and Pathology of the Skin.* Eds., Rook, A. J. and Walton, G. S., F. A. Davis, Co., Philadelphia, 1965.
10. Fitzpatrick, T. B., Brunet, P. and Kukita, A.: The nature of hair pigment, p. 255, *The Biology of Hair Growth.* Eds., Montagna, W. and Ellis, R. A., Academic Press, New York, 1958.
11. Taylor, A. C.: Survival of rat skin and changes in hair pigmentation following freezing. J. Exp. Zool., *110:* 77, 1949.
12. Mikhail, G. R.: Hair neogenesis in rat skin. Arch. Derm., *88:* 713, 1963.
13. Billingham, R. E.: A reconsideration of the phenomenon of hair neogenesis with particular reference to the healing of cutaneous wounds in adult mammals, p. 451, *The Biology of Hair Growth.* Eds., Montagna, W. and Ellis, R. A., Academic Press, New York, 1958.
14. Sanford, B. H., Chase, H. B., Carroll, S. B. and Arsenault, C. T.: The effects of grafting during various stages of the hair growth cycle. Anat. Record, *152:* 17, 1965.

15. Chase, H. B.: Induced melanocyte deactivation and reactivation in hair follicles, p. 503, *The Pigmentary System.* Eds., Montagna, W. and Hu, F., Pergamon Press, New York, 1966.
16. Sato, C. and Sakka, M.: Depigmentation of hair in C57 BL mice after injection of irradiated thymic RNA. J. Exp. Med., *93:* 23, 1967.
17. Kostanecki, W., Kwiatkowska, E. and Gawkowska, M.: The effect of inflammatory conditions of the scalp upon hair growth and hair melanogenesis in man. Dermatologica, *136:* 95, 1967.
18. Lyne, A. G., Hollis, D. E., Chase, H. B. and Hayman, R. H.: Changes experimentally produced in the pigmentation of the skin and coat of sheep. Aust. J. Sci., *30:* 30, 1967.
19. Munan, L. P.: Depigmentation of hair by oxidized adrenalin solutions. Lancet, *1:* 600, 1953.